

WO9321913

Publication Title:

ANTI-OBESITY DRUGS

Abstract:

Obesity is treated by the administration to a subject of a compound having the general formula (I): $R_4-(CH_2)_n-CO-N(R_1)-CH(R_2)-CO(-R_3)$, wherein R_1 represents H or CH_3 ; R_2 represents a side chain of a naturally occurring amino acid; R_3 represents OH, OCH_2CH_3 and NH_2 ; n is 6-18; and R_4 represents CH_3 or a group having the general formula (II): $R_3-CO-CH(R_2)-N(R_1)-CO-$, wherein R_1 , R_2 and R_3 have the above meanings. The compounds of formula (I) wherein R_4 is a group of formula (II), are novel compounds.

Data supplied from the esp@cenet database - <http://ep.espacenet.com>



INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification ⁵ : A61K 31/225, 31/195 C07C 233/47, 237/22	A1	(11) International Publication Number: WO 93/21913 (43) International Publication Date: 11 November 1993 (11.11.93)
(21) International Application Number: PCT/EP93/01014 (22) International Filing Date: 27 April 1993 (27.04.93) (30) Priority data: 101708 28 April 1992 (28.04.92) IL (71) Applicants (for all designated States except US): SENYORINA LTD. [IL/IL]; 8 Hashoshanim Street, 45 421 Herzlia (IL). COHN, Michael [DE/IL]; 21 Hazorea Street, 46 910 Kfar Shmaryahu (IL). (72) Inventor; and (75) Inventor/Applicant (for US only) : SHINITZKY, Meir [IL/IL]; 20 Derech Haganim, 46 910 Kfar Shmaryahu (IL). (74) Agent: VOSSIUS & PARTNER; Siebertstr. 4, D-8000 Munich 80 (DE).		(81) Designated States: AT, AU, BB, BG, BR, CA, CH, CZ, DE, DK, ES, FI, GB, HU, JP, KP, KR, LK, LU, MG, MN, MW, NL, NO, NZ, PL, PT, RO, RU, SD, SE, SK, UA, US, European patent (AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG). Published <i>With international search report.</i>
(54) Title: ANTI-OBESITY DRUGS (57) Abstract Obesity is treated by the administration to a subject of a compound having the general formula (I): $R_4-(CH_2)_n-CO-N(R_1)-CH(R_2)-CO(-R_3)$, wherein R_1 represents H or CH_3 ; R_2 represents a side chain of a naturally occurring amino acid; R_3 represents OH, OCH_2CH_3 and NH_2 ; n is 6-18; and R_4 represents CH_3 or a group having the general formula (II): $R_3-CO-CH(R_2)-N(R_1)-CO-$, wherein R_1 , R_2 and R_3 have the above meanings. The compounds of formula (I) wherein R_4 is a group of formula (II), are novel compounds.		

FOR THE PURPOSES OF INFORMATION ONLY

Codes used to identify States party to the PCT on the front pages of pamphlets publishing international applications under the PCT.

AT	Austria	FR	France	MR	Mauritania
AU	Australia	GA	Gabon	MW	Malawi
BB	Barbados	GB	United Kingdom	NL	Netherlands
BE	Belgium	GN	Guinea	NO	Norway
BF	Burkina Faso	GR	Greece	NZ	New Zealand
BG	Bulgaria	HU	Hungary	PL	Poland
BJ	Benin	IE	Ireland	PT	Portugal
BR	Brasil	IT	Italy	RO	Romania
CA	Canada	JP	Japan	RU	Russian Federation
CF	Central African Republic	KP	Democratic People's Republic of Korea	SD	Sudan
CG	Congo	KR	Republic of Korea	SE	Sweden
CH	Switzerland	KZ	Kazakhstan	SK	Slovak Republic
CI	Côte d'Ivoire	LI	Liechtenstein	SN	Senegal
CM	Cameroon	LK	Sri Lanka	SU	Soviet Union
CS	Czechoslovakia	LU	Luxembourg	TD	Chad
CZ	Czech Republic	MC	Monaco	TG	Togo
DE	Germany	MG	Madagascar	UA	Ukraine
DK	Denmark	ML	Mali	US	United States of America
ES	Spain	MN	Mongolia	VN	Viet Nam
FI	Finland				

ANTI-OBESITY DRUGS

FIELD OF THE INVENTION

The present invention is in the field of treatment and prevention of obesity. The present invention provides compositions and methods for the treatment or prevention of such disorders utilizing, as active ingredient, lipophilic derivatives of natural amino acids. Furthermore, the present invention provides certain such novel compounds.

PRIOR ART

The following prior art is believed to be relevant as a background to the present invention:

1. Bar-Tana *et al.*, *J. Biol. Chem.*, 1985, 260, 8404-8410.
2. Rose-Kahn *et al.*, *J. Biol. Chem.*, 1985, 260, 8411-8415.
3. Bar-Tana *et al.*, *J. Lipid Res.*, 1988, 29, 431-441.
4. Frenkel *et al.*, *J. Biol. Chem.*, 1988, 263, 8491-8497.
- 15 5. Tzur *et al.*, *Diabetes*, 1988, 37, 1618-1624.
6. U.S. Patent No. 4,634,795.
7. U.S. Patent No. 4,689,344.
8. U.S. Patent No. 4,711,896.
9. U.S. Patent No. 4,908,385.

In the following text, reference to these prior art publications will be made by indicating in brackets their number from the above list.

BACKGROUND OF THE INVENTION

5 Energy from food is primarily provided by carbohydrates and lipids. Carbohydrates usually supply the immediate energy needs and their excess is stored as glycogen in the liver or converted to lipids. Lipids can also be metabolized as immediate energy providing substances but their rate of energy provision is relatively slow and they are generally stored in the
10 body for use in states of deprivation. Lipid is stored in the body mostly as fat under the skin and consumption of lipids and carbohydrates beyond the metabolic need leads to fattening. The associated medical and aesthetic problems, are a major concern in modern society.

Apart from surgery and dietary means for reduction of fat
15 absorption in the small intestine, there are presently no satisfactory means for reducing fat storage in the body and the current means of choice are still diet and exercise. There is, however, a desire for drugs which will reduce fat accumulation by inhibiting lipid and lipoprotein synthesis in the liver. Recently, a series of β,β' tetramethyl substituted α,ω dicarboxylic acids
20 (MEDICA) have been synthesized and suggested as potential anti-fattening drugs^(1-4,6-9). The most potent drug of this series was found to be the hexadecane derivative (MEDICA 16). It was demonstrated that MEDICA, which is a non-naturally occurring fatty acid, could inhibit biosynthetic pathways of triglycerides and cholesterol in the liver. Experiments with
25 MEDICA 16 given in the diet to normal and obese rats have indicated a strong inhibition of glyceride and cholesterol biosynthesis in the liver evidenced by a marked reduction in their serum contents⁽³⁾. Furthermore, in the obese animals adipose tissue was reduced by about 75% over the whole body concomitantly to extensive weight loss⁽⁵⁾. However, the metabolic

clearance of these compounds via integration into glycerol esters or via oxidation is relatively slow due to the presence of carboxylate at the two edges of the molecules and the β alkyl substitution. Despite their impressive effect, MEDICA are expected to exert a long term toxicity due to their non-compatible molecular structure. Thus, chronic intake of MEDICA, which is required for maintaining a low fat state, would likely be associated with adverse toxic effects in the long run.

OBJECTS OF THE INVENTION

It is the object of the present invention to provide a pharmaceutical composition, method and dietary supplements for the treatment and/or prevention of obesity. More specifically, it is an object of the present invention to provide such composition and method utilizing lipophilic derivatives of natural amino acids.

It is a further object of the present invention to provide certain novel lipophilic derivatives of natural amino acids useful in such compositions and methods.

The remaining objects of the present invention will be illustrated from the following description and claims.

GENERAL DESCRIPTION OF THE INVENTION

By a first of its aspects, the present invention provides a pharmaceutical composition for the treatment of obesity comprising a pharmaceutically acceptable carrier and, as an active ingredient, a compound having the general Formula I:



wherein R_1 represents H or CH_3 ;
 R_2 represents a side chain of a naturally occurring amino acid;
 R_3 represents OH, OCH_2CH_3 and NH_2 ;
 n is 6 - 18, preferably 12 - 16; and
 R_4 represents CH_3 or a group having the general Formula II:



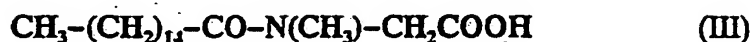
wherein R_1 , R_2 and R_3 have the above meanings.

10 The present invention also provides methods for the treatment of obesity comprising administering to a subject in need an effective amount of an active ingredient being a compound having the general Formula I as defined above.

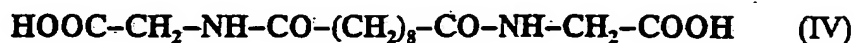
15 The present invention also provides a dietary supplement for the prevention of obesity comprising an active ingredient being a compound of the general Formula I as defined above.

The compounds according to Formula I wherein R_4 represents a group of Formula II are novel, and such compounds are also provided by the present invention.

20 An example of compounds of Formula I wherein R_4 is CH_3 , is N-palmitoyl sarcosine (P-Sar) having the Formula III:



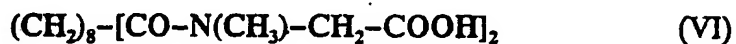
25 Examples of compounds of Formula I, in which R_4 represents a group having the general Formula II, are N,N' sebacoyl bis-glycine (GSG), having the Formul IV:



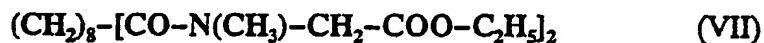
N,N' sebacoyl bis *l*-aspargin (NSN), having the Formula V:



5 N,N' sebacoyl bis-sarcosine [S(Sar)₂] having the Formula VI:

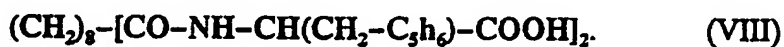


10 N,N' sebacoyl bis-sarcosine-ethylester [S(SarOEt)₂], having the Formula VII:



and

15 N,N' sebacoyl bis-phenylalanine (FSF), having the Formula VIII:



BRIEF DESCRIPTION OF THE DRAWINGS

20 Fig. 1 shows the effect of NSN (0.1% w/w in purina) on weight of 3 months old mice during a 1 month *ad lib.* feeding;

Fig. 2 shows the effect of several anti-fattening agents including a fattening diet in accordance with the invention (0.1% w/w in purina + 6% corn oil) on weight of 3 months old mice;

25 Fig. 3 shows the daily weight of adult mice administered with various anti-fattening agents of the invention; and

Fig. 4 shows the effect of treatment with NSN on the ³H₂O incorporation to liver and adipose tissue of adult mice.

The present invention will now be described with reference to the following non-limiting examples.

Example 1: preparation of compounds

5 Lypophilic derivatives of the natural amino acids can be synthesized via the N-hydroxy succinimide ester of the respective fatty acid. Such active esters react with primary or secondary amine to form the corresponding amide while liberating a free N-hydroxy succinimide.

In the following are three examples of such synthesis:

10

A: preparation of P-Sar

Hydroxy succinimide ester of palmitic acid (PHS) was obtained from SIGMA. One volume of 30mM PHS in tetrahydrofuran was mixed with one volume of excess sarcosine (0.3 M) in 0.1 M of aqueous sodium
15 bicarbonate according to the procedure described by Lapidot *et al*, *J. Lipids Res.*, 1967, 8, 142. The mixture was mixed at 40°C for 24 hours. The tetrahydrofuran was then evaporated and the mixture was acidified to pH 1 with HCl, whereupon the crude product precipitated and was collected. After washing with water the product was crystallized from isopropanol.

20

B: preparation of GSG

(a) 1 mole of sebacic acid was reacted with 2 moles of N-hydroxysuccinimide and 2 moles of dicyclohexyl carbodiimide in ethyl acetate. The resulting compound, 1,10 sebacoyl di (N-hydroxy-succinimide) ester, [Seb-(NHS)₂] was crystallized from isopropanol.
25 Seb(NHS)₂ was found to have an m.p. of 159°C.

(b) 1 volume of 30mM Seb(NHS)₂ in tetrahydrofuran with 1 volume excess glycine (0.3 M) in aqueous 0.1 M sodium bicarbonate

according to the procedure described by Lapidot *et al.*, *supra*. After 24 hours of mixing at 40°C, the tetrahydrofuran was evaporated under reduced pressure and the product was precipitated by acidifying with 1M HCl to pH 1. The precipitate was collected and washed with water. Crystallization
5 was from isopropanol.

C: preparation of NSN

NSN was prepared by the same procedure of Example (B) except that instead of glycine in step (b), 1-asparagine was used.
10

The remaining compounds described in the following were prepared in a similar manner, *mutatis mutandis*.

Example 2: Experimental results

15 (a) Two groups of 5 three months old mice were fed with purina, *ad lib*. The diet of 1 group was supplemented with NSN (0.1% w/w in purina). The weight increase of the mice in each group was measured over 31 days and the results shown in Fig. 1 clearly demonstrate that the weight increase of the experimental group, was far less than that in the control
20 group.

(b) 5 groups of 5 three months old mice each, were fed with a fattening diet consisting of purina and 6% corn oil, and out of these five groups, the diet of four was supplemented with anti-fattening drugs in accordance with the invention (0.1% w/w in the food). The following drugs
25 were tested: P-Sar, GSG, S(Sar)₂ and S(SarOEt)₂.

The results shown in Fig. 2 clearly demonstrate that the weight increase of the treated animals was far less than that of the animals of the control group.

(c) Adult mice were divided into 9 groups of five mice each, and were fed with normal purina *ad lib.* and 8 groups received one of the following supplements in their diet (0.25% or 0.35% w/w in the food): dNSN, NSN, dFSF and FSF.

5 One group did not receive any supplement and served as control.

Food consumption was *ad lib.*

The treatment was over a period of 40 days after which it was ceased and all groups of animals returned to a normal diet.

10 The results shown in Fig. 3 clearly demonstrate that some of the supplements caused even a slight increase over control. Thus for example, while an increase was observed with 0.25% FSF, a considerable decrease in weight over the entire tested period was observed with 0.35% FSF. Accordingly it is believed that upon increase of the concentration of
15 these drugs they will all have an anti-fattening affect.

(d) 8-12 months old mice were administered with NSN either intra peritonelly (I.P.) or Per Os (P.O.), 10 mg per day for 4 days. The incorporation of $^3\text{H}_2\text{O}$ to liver and adipose tissue was tested. For that purpose tritiated water was injected I.P. after overnight fast and 2 hours later
20 the animals were sacrificed and the incorporation into lipids of the liver and the adipose tissue were determined by measuring radioactivity.

The results shown in Fig. 4, clearly demonstrate the decrease in treated water incorporation into the treated animals.

CLAIMS:

1. A pharmaceutical composition for the treatment of obesity comprising a pharmaceutically acceptable carrier and, as an active ingredient, a compound having the general Formula I:

5



wherein R_1 represents H or CH_3 ;

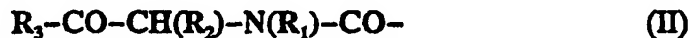
R_2 represents a side chain of a naturally occurring amino acid;

10

R_3 represents OH, OCH_2CH_3 and NH_2 ;

n is 6 - 18; and

R_4 represents CH_3 or a group having the general Formula II:



15

wherein R_1 , R_2 and R_3 have the above meanings.

2. A pharmaceutical composition according to Claim 1, comprising as active ingredient a compound which is a member of the group consisting of N-palmitoyl sarcosin, N,N'-sebacoyl bis-glycine, N',N'-sebacoyl bis ℓ -asparagine, N,N'-sebacoyl bis d-asparagine N',N'-sebacoyl bis sarcosin, N,N'-sebacoyl bis-sarcosin-ethylester and N,N'-sebacoyl bis-phenylalanine.

20

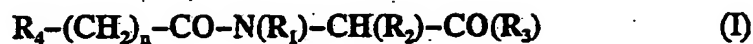
3. A dietary supplement for preventing obesity comprising a compound of formula I as defined in Claim 1.

25

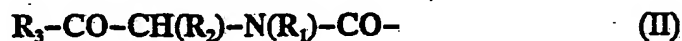
4. A method for the treatment of obesity comprising administering to a subject in need an effective amount of a compound of formula I as defined in Claim 1.

5. Use of a compound having the general formula I as defined in claims 1 or 2 for the preparation of a pharmaceutical composition for the treatment of obesity.

6. A compound of the general formula I:



wherein R_1 represents H or CH_3 ;
 R_2 represents a side chain of a naturally occurring amino acid;
 R_3 represents OH, OCH_2CH_3 and NH_2 ;
 n is 6 - 18; and
 R_4 represents a group of the general formula II:



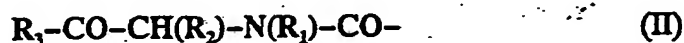
wherein R_1 , R_2 and R_3 have the above meanings.

7. A pharmaceutical composition comprising a pharmaceutically acceptable carrier and, as an active ingredient, a compound according to claim 6.

8. Use of a compound having the general Formula I:



wherein R_1 represents H or CH_3 ;
 R_2 represents a side chain of a naturally occurring amino acid;
 R_3 represents OH, OCH_2CH_3 and NH_2 ;
 n is 6 - 18; and
 R_4 represents CH_3 or a group having the general Formula II:



wherein R_1 , R_2 and R_3 have the above meanings for combatting obesity.

1/4

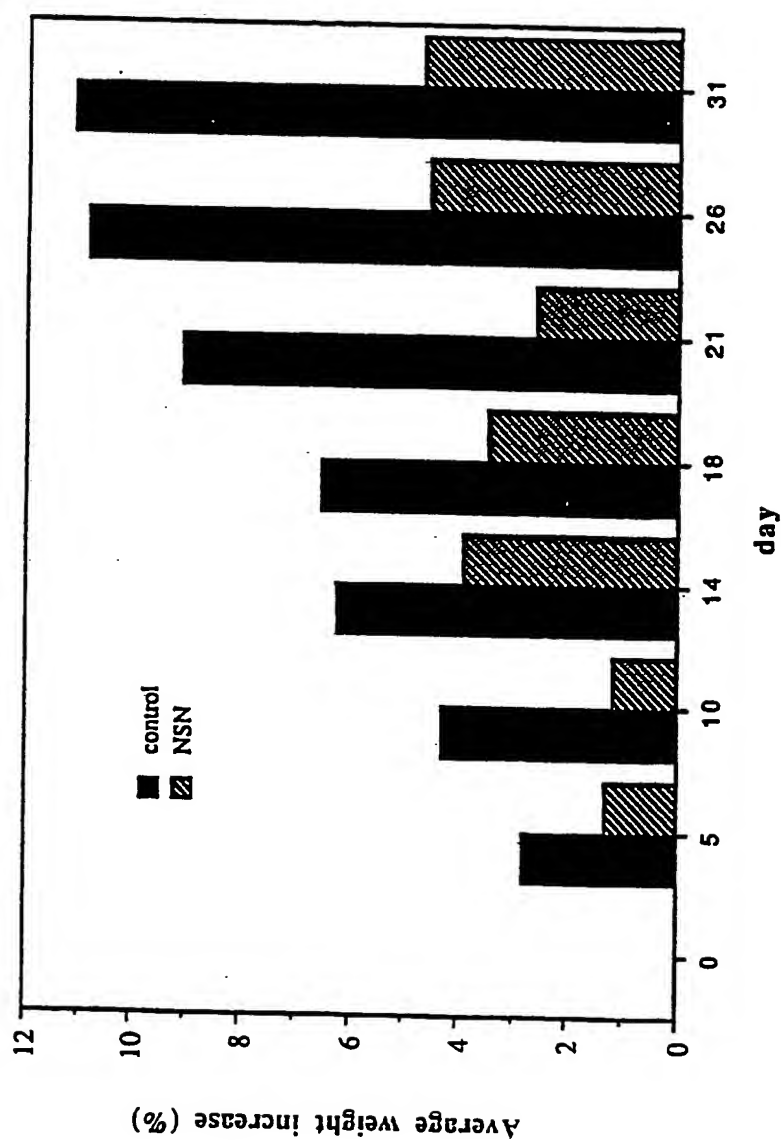


Figure 1

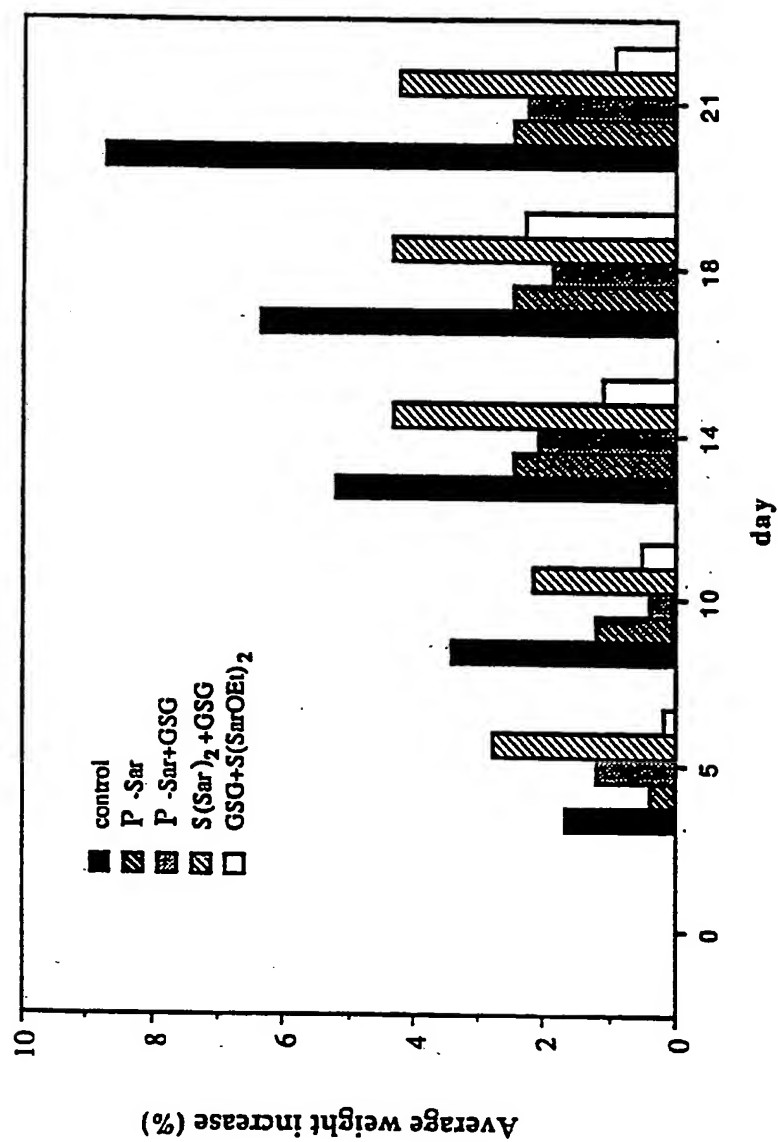


Figure 2:

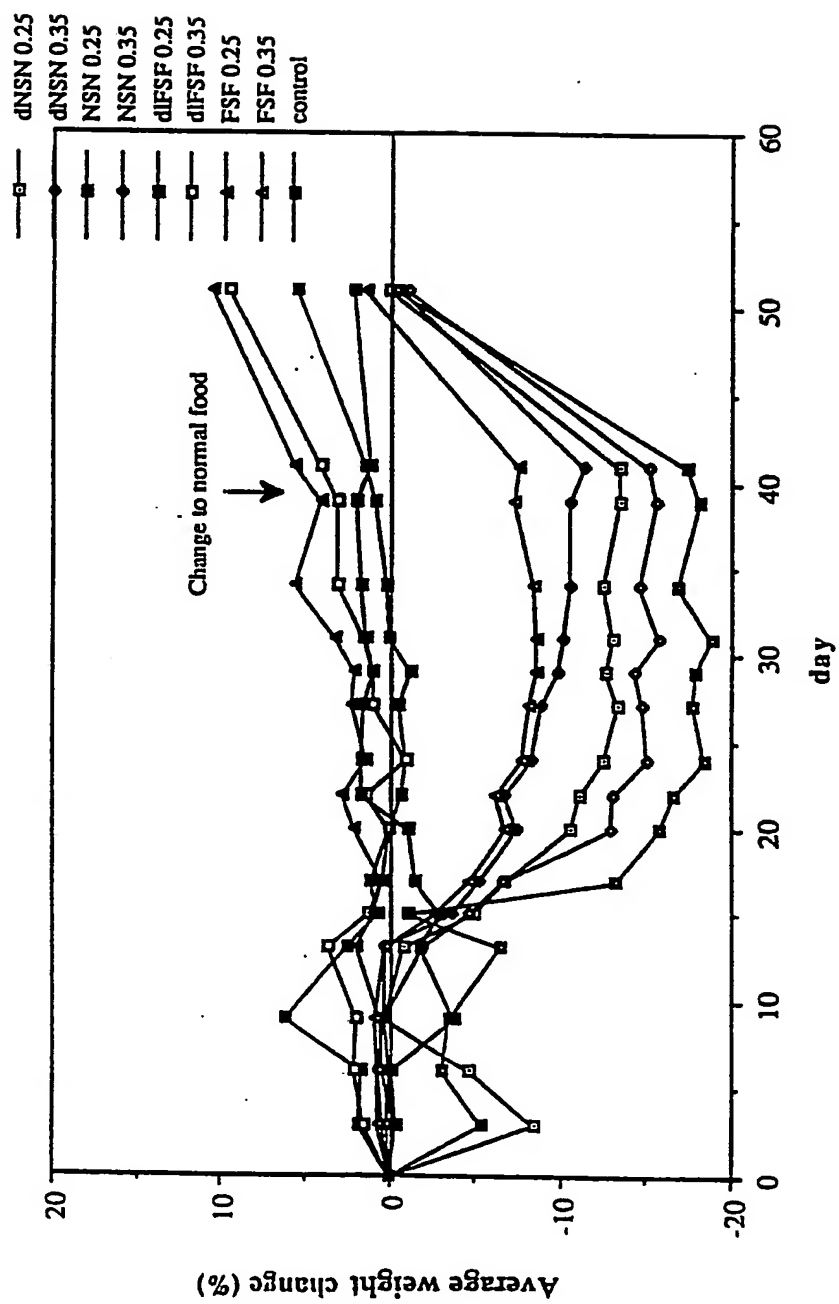


Figure 3:

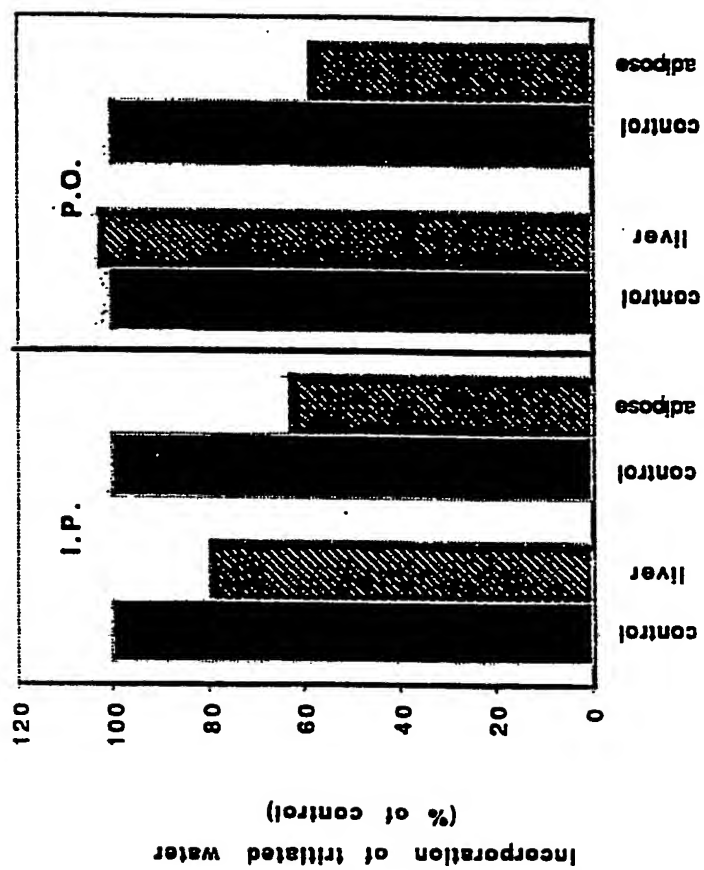


Figure 4:

INTERNATIONAL SEARCH REPORT

PCT/EP 93/01014

International Application

I. CLASSIFICATION OF SUBJECT MATTER (If several classification symbols apply, indicate all) ⁶		
According to International Patent Classification (IPC) or to both National Classification and IPC		
Int.Cl. 5 A61K31/225; A61K31/195; C07C233/47; C07C237/22		
II. FIELDS SEARCHED		
Minimum Documentation Searched ⁷		
Classification System	Classification Symbols	
Int.Cl. 5	A61K ; C07C	
Documentation Searched other than Minimum Documentation to the Extent that such Documents are Included in the Fields Searched ⁸		
III. DOCUMENTS CONSIDERED TO BE RELEVANT⁹		
Category ¹⁰	Citation of Document, ¹¹ with indication, where appropriate, of the relevant passages ¹²	Relevant to Claim No. ¹³
X	DE,A,2 234 399 (DR. KARL THOMAE GMBH) 31 January 1974 see page 9 see claims 1,2	1,2
X	--- J. CHEM. SOC., CHEM. COMMUN. no. 9, 1986, pages 659 - 661 R.T.C. BROWNLEE 'The synthesis and characterization of a series of bis-intercalating bis-anthracyclines' see page 659 --- -/-	6
<p>¹⁰ Special categories of cited documents : ¹⁰</p> <p>"A" document defining the general state of the art which is not considered to be of particular relevance</p> <p>"E" earlier document but published on or after the international filing date</p> <p>"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)</p> <p>"O" document referring to an oral disclosure, use, exhibition or other means</p> <p>"P" document published prior to the international filing date but later than the priority date claimed</p> <p>"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention</p> <p>"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step</p> <p>"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.</p> <p>"A" document member of the same patent family</p>		
IV. CERTIFICATION		
Date of the Actual Completion of the International Search	Date of Mailing of this International Search Report	
30 JULY 1993	11. 08. 93	
International Searching Authority	Signature of Authorized Officer	
EUROPEAN PATENT OFFICE	KRAUTBAUER B.	

III. DOCUMENTS CONSIDERED TO BE RELEVANT (CONTINUED FROM THE SECOND SHEET)		
Category ^a	Citation of Document, with indication, where appropriate, of the relevant passages	Relevant to Claim No.
X	<p>KHIM.-FARM. ZH. vol. 26, no. 2, 1992, pages 43 - 45 N.A. GRIGORYAN 'Synthesis and antistaphylococcal activity of dicarboxylic acid derivatives containing an amino acid fragment' see page 44 see abstract</p>	1,2,6,7
A	<p>US,A,4 711 896 (J. BAR-TANA ET AL.) 8 December 1987 cited in the application see column 1 see claims 1-8</p>	1-8
A	<p>DIABETES vol. 37, 1988, pages 1618 - 1624 R. TZUR ET AL. 'Hypolipidemic, antiobesity, and hypoglycemic-hypoinsulinemic effects of beta,beta'-methyl-substituted hexadecanedioic acid in sand rats' cited in the application see the whole document</p>	1-8

INTERNATIONAL SEARCH REPORT

International application No.

/EP 93/01014

Box I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This international search report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☒ **Claims Nos.:**
because they relate to subject matter not required to be searched by this Authority, namely:
REMARK: Although claims 4 and 8 are directed to a method of treatment of the human/animal body the search has been carried out and based on the alleged effects of the compound/composition.
2. ☒ **Claims Nos.:**
because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:
Claims searched completely: 2. Claims searched incompletely: 1,3-8.
Due to the large number of compounds which are theoretically defined by the markush formulas of claims 1,6 and 8, the search had to be restricted to the compounds explicitly mentioned in the description and the claims.
3. ☐ **Claims Nos.:**
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

1. ☐ As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.
2. ☐ As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.
3. ☐ As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.:
4. ☐ No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

- ☐ The additional search fees were accompanied by the applicant's protest.
- ☐ No protest accompanied the payment of additional search fees.

ANNEX TO THE INTERNATIONAL SEARCH REPORT ON INTERNATIONAL PATENT APPLICATION NO.

EP 9301014
SA 73398

This annex lists the patent family members relating to the patent documents cited in the above-mentioned international search report.
The members are as contained in the European Patent Office EDP file on
The European Patent Office is in no way liable for these particulars which are merely given for the purpose of information.

30/07/93

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
DE-A-2234399	31-01-74	AT-B- 338434	25-08-77
		AU-A- 5812973	16-01-75
		BE-A- 802414	16-01-74
		CA-A- 1022849	20-12-77
		JP-A- 49085244	15-08-74
		NL-A- 7309851	21-01-74
US-A-4711896	08-12-87	DE-A- 3423166	02-01-86
		AU-A- 4607185	24-01-86
		CA-A- 1262552	31-10-89
		WO-A- 8600298	16-01-86
		EP-A, B 0185080	25-06-86
		JP-T- 61502537	06-11-86

**This Page is Inserted by IFW Indexing and Scanning
Operations and is not part of the Official Record**

BEST AVAILABLE IMAGES

Defective images within this document are accurate representations of the original documents submitted by the applicant.

Defects in the images include but are not limited to the items checked:

☐ BLACK BORDERS

☒ IMAGE CUT OFF AT TOP, BOTTOM OR SIDES

☐ FADED TEXT OR DRAWING

☐ BLURRED OR ILLEGIBLE TEXT OR DRAWING

☐ SKEWED/SLANTED IMAGES

☐ COLOR OR BLACK AND WHITE PHOTOGRAPHS

☐ GRAY SCALE DOCUMENTS

☐ LINES OR MARKS ON ORIGINAL DOCUMENT

☒ REFERENCE(S) OR EXHIBIT(S) SUBMITTED ARE POOR QUALITY

☐ OTHER: _____

IMAGES ARE BEST AVAILABLE COPY.

As rescanning these documents will not correct the image problems checked, please do not report these problems to the IFW Image Problem Mailbox.